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MICROBIAL DECONTAMINATION AND SAMPLING PROGRAM FOR ANCHORED INTERPLANETARY MONITORING PLATFORM (AIMP-E) SPACECRAFT

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GODDARD SPACE FLIGHT CENTER
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ABSTRACT

A requirement of the Office of Planetary Quarantine, NASA Headquarters for biologically clean spacecraft operating in the near vicinity of the moon necessitated the development of a decontamination program for the AIMP-E spacecraft.

Decontamination was effected with chemical solutions of isopropyl alcohol and acetone. To determine effectiveness of decontamination process two methods of recovering viable micro-organisms were used. One method employed control strips with detachable coupons for monitoring the electronic circuit modules and the other method employed sterile swabs and templates to monitor other surface areas. Coupons and/or swabs were immersed in a 1% peptone wash solution and sonicated at 25 kc/sec for 12 minutes. Aliquots of the sonicated solution were plated out on agar, incubated and colony counts made. Records were made of the numbers of aerobic and anaerobic spore and vegetative organisms remaining on a surface after the decontamination process. All assembly and test operations were conducted in controlled and/or clean-room facilities. As a result of the evaluation of records that were maintained the AIMP-E spacecraft contained an internal burden of 2.19×10^5 organisms and a surface burden of 4.42×10^4. The surface spore loading was estimated at 7.4×10^3 organisms. This spore population on the surface will be reduced to less than 1.89×10^{-9} due to the spacecraft's orbital life expectancy of 3 years and 1440 cycles of temperature change in an ultra high vacuum.

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MICROBIAL DECONTAMINATION AND SAMPLING PROGRAM FOR ANCHORED INTERPLANETARY MONITORING PLATFORM (AIMP-E) SPACECRAFT

INTRODUCTION

A primary objective of the Anchored Interplanetary Monitoring Platform (AIMP-E) is to investigate the characteristics of the interplanetary magnetic field out to and at lunar distances in either a captured lunar orbit or a geocentric orbit with an apogee near or beyond the lunar distance. Because of the spacecraft's mission in the near vicinity of the moon it is considered a potential lunar lander as the gravitational pull of the moon will eventually capture the spacecraft and it will impact the lunar surface. When this occurs a number of viable micro-organisms remaining on the spacecraft carried from the earth will be deposited on the lunar surface.

It being a prime responsibility of the NASA Bioscience program to record and maintain inventories of all biological contamination deposited on the lunar surface it then became necessary for the Goddard Space Flight Center (GSFC) to device an in-house program for biological decontamination; sampling and assaying the spacecraft and its hardware, maintaining a bio-clean environment during the phases of assembly and testing and a method of maintaining running records of viable contamination.

Before a program for biological decontamination of the AIMP-E spacecraft was inaugurated tests were conducted to determine the compatibility of the spacecraft's materials and the decontaminates used to reduce the micro-biological population. Samples of all materials used in the build-up of the spacecraft system were tested, i.e., all metals coated and bare, fiberglass, epoxies, sealants, adhesives, thermal coatings, plastics and working electronic components. As a result of all the tests conducted there was no evidence to substantiate that failure of a component would occur as a result of the decontamination process.

All surfaces of the spacecraft were monitored for microbiological contamination. The various areas were classified with respect to the manner in which they were exposed or occluded and as to their physical location. Figure 1 depicts the AIMP-E spacecraft in a flight configuration.

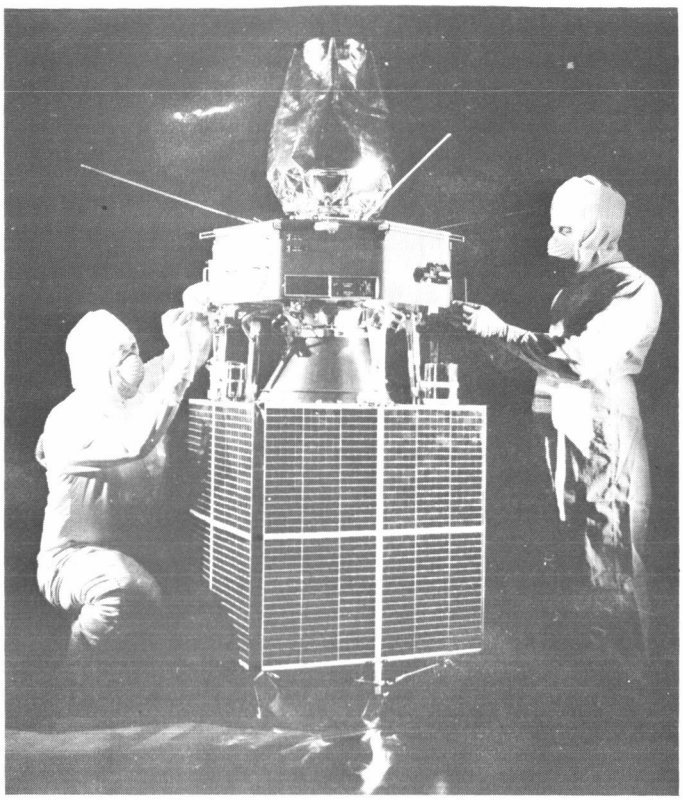

Figure 1. AIMP-E Spacecraft in a Flight Configuration

AREA CLASSIFICATION

A. Interior surfaces prior to occlusion by conformal coating and/or encapsulation. All interior surface areas of the module frames which included walls, cavities, and electronic circuit boards.

B. Occluded surface areas obstructed by module frames excluding exposed surfaces of module frame stacks.

C. Cover and occluded inner surfaces of the spacecraft.

D. Other interior surfaces of the spacecraft.

E. Exterior surfaces of the spacecraft occluded and exposed.

METHODS EMPLOYED FOR MICROBIAL RECOVERY

Two methods of recovering viable organisms from the spacecraft surfaces were employed. One method employed control strips with detachable coupons. Figure 2 shows a control strip in detail. Figure 3 shows such a control strip affixed to a circuit module frame. The control strips were first sterilized in a

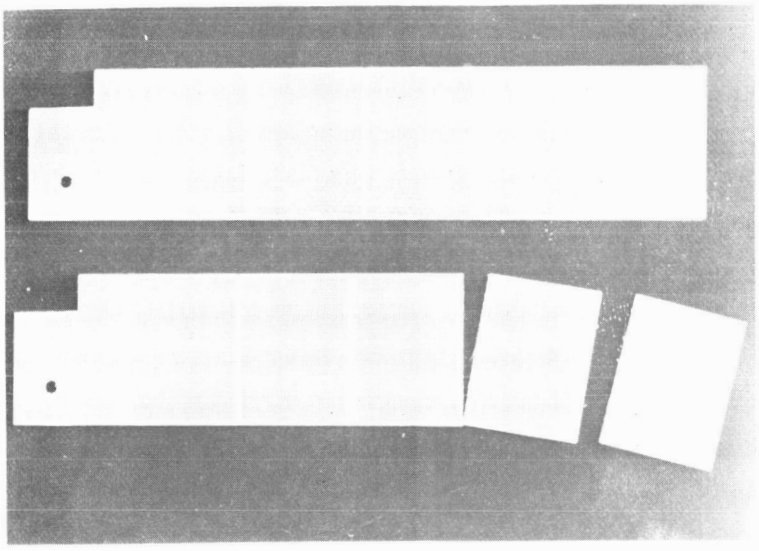

Figure 2. Control Strip with Detachable Coupons

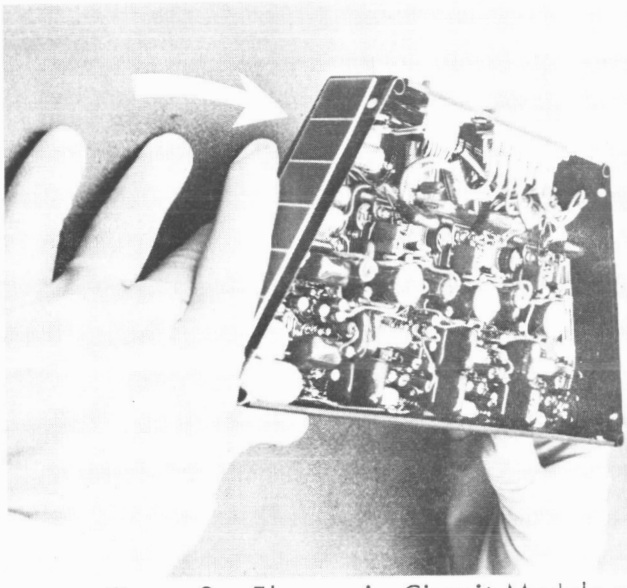

Figure 3. Electronic Circuit Module with Control Strip Attached

steam autoclave then affixed to each module frame in a similar manner, so located as to compel the technician to touch the control strip each time the module is handled. The purpose of the control strip is to prove the effectiveness of decontamination process and allow a practical method of assaying the contamination level on a circuit module prior to decontamination and the probable level of contamination after the decontamination process. These control strips were fabricated from the same material as the printed circuit board and in such a manner as to yield five easily removed coupons. A coupon was removed from the control strip and assayed to determine the contamination level. After the decontamination process another coupon was removed so as to determine the number of viable organisms remaining on the circuit module. Upon removing a coupon it was placed into a wash bottle containing 50 ml of 1.0% sterile peptone solution. The wash bottle was then placed in an ultrasonic bath and sonicated at 25 kc/sec for 12 minutes before petri-plate preparation.

The circuit modules were monitored biologically in this manner at time of conformal coating and again at time of encapsulation.

The second method of recovering viable micro-organisms employed sterile swabs and templates. Sampling was accomplished by first inserting the sterile swab into the wash solution. The swab was then retracted into its holder and the excess solution removed by pressing the swab tip on the inside wall of the tube. A rolling twisting motion was used. Sterile Kraft paper was used as a template. The template openings varied depending upon the configurations of areas sampled. In most cases a 4 square inch opening template was used. After sampling a particular area the microbiologist performing the sampling held the swab by its protective shield and aseptically broke it off into its test tube which contained 5 ml of sterile 1.0% peptone wash solution. The test tube containing the swab was then placed into an ultrasonic bath and sonicated at 25 kc/sec for 12 minutes. This procedure dispensed the cotton into the peptone solution. Figure 4 shows the type of swab used to obtain the samples from the spacecraft and its hardware.

Petri-Plate Preparation

Eight, 100 mm-diameter petri plates were prepared from each coupon wash bottle. Four, (4) plates were prepared each containing 5 ml of the sonicated solution and 20 ml of trypticase soy agar (TSA). The remaining sonicated solution was heat-shocked at 80°C in a water bath for 20 minutes. Four, (4) petri plates were prepared each containing 5 ml of the heat-shocked solution. Four, (4) petri plates were prepared from each swab sample. Two, (2) plates were prepared each containing 1 ml of solution and 20 ml of TSA. Remaining solution was heat-shocked at 80°C for twenty minutes. Two, (2) plates were then prepared each containing 1 ml of the heat-shocked solution.

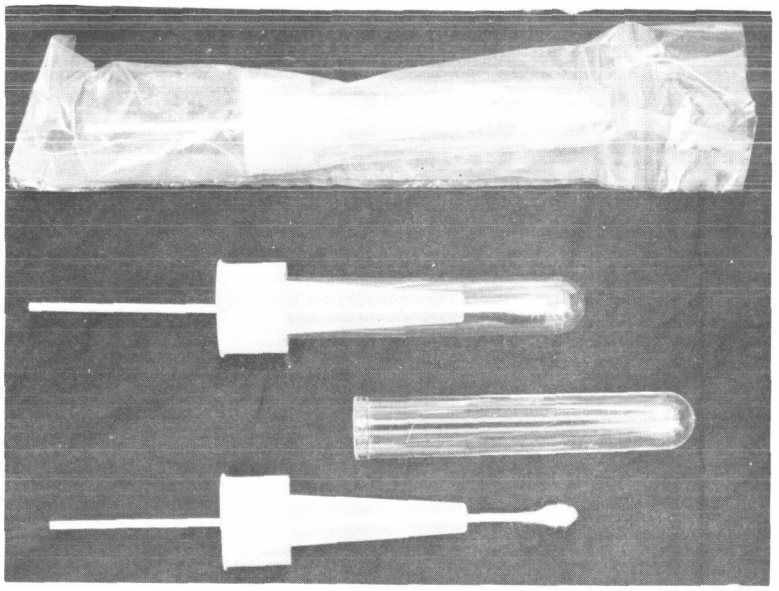

Figure 4. Sterile Swabs Used for Sampling Areas for Assays

Culturing

Two (2) petri plates prepared from the non-heat shocked solution obtained from the coupons and two, (2) plates prepared from the heat-shocked portion of this solution were aerobically incubated. Colony counts were made after 24, 48 and 72 hours. Remaining plates containing nonheat-shocked and heat-shocked portions of solution were anaerobically incubated. Colony counts were made after 72 hours of incubation. Culturing of the solution obtained from the swab samples were performed in the same manner. All petri plates were incubated at 32°C. Culturing normally was started within an hour after taking samples from the spacecraft. Figures 5a and 5b depict the manner of culture preparation.

All surfaces of the spacecraft were monitored for microbial contamination, i.e., the areas which would be occluded by an attachment, instrument or structural member and the exposed surface areas on the interior and exterior of the spacecraft. Figure 6 is a schematic that depicts the spacecraft configuration without its 4th stage retromotor, attitude control system, nutation damper and solar paddles. It also depicts the structural materials.

Figure 7 depicts the manner in which the spacecraft and the various components that make up the spacecraft system were decontaminated and the manner in which the microbial samples were taken.

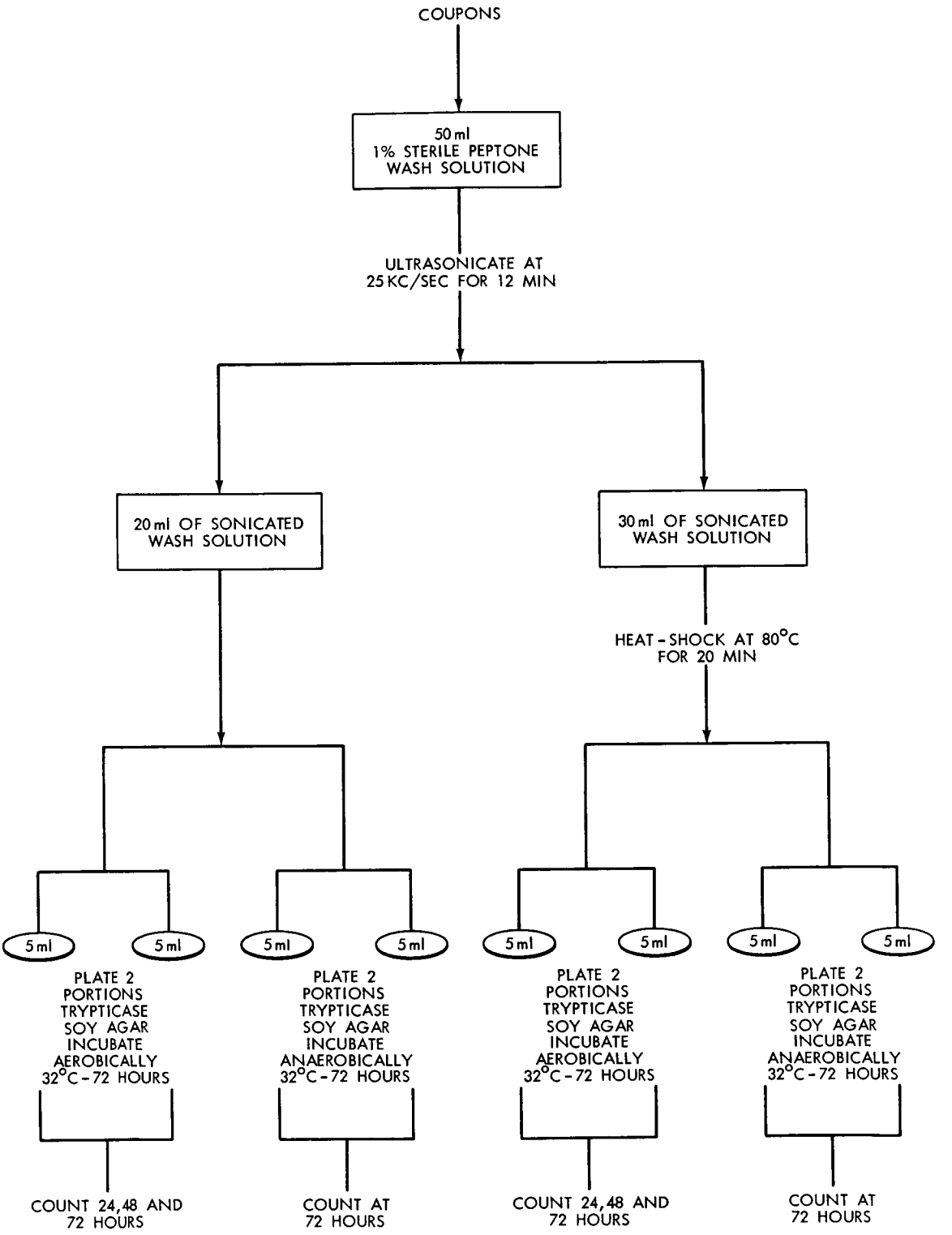

Figure 5a. Schematic of Coupon Assaying Procedure

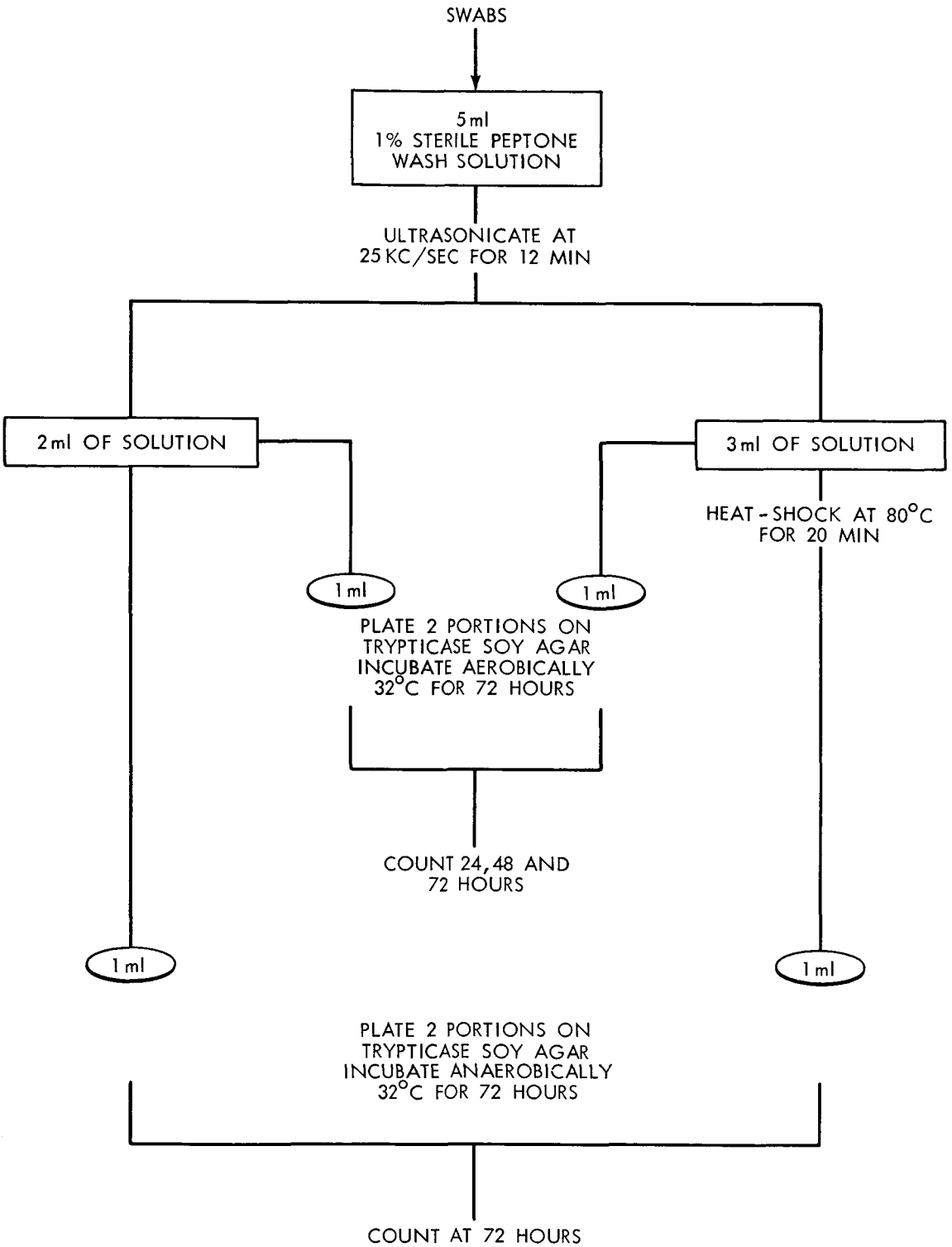

Figure 5b. Schematic of Swab Assaying Procedure

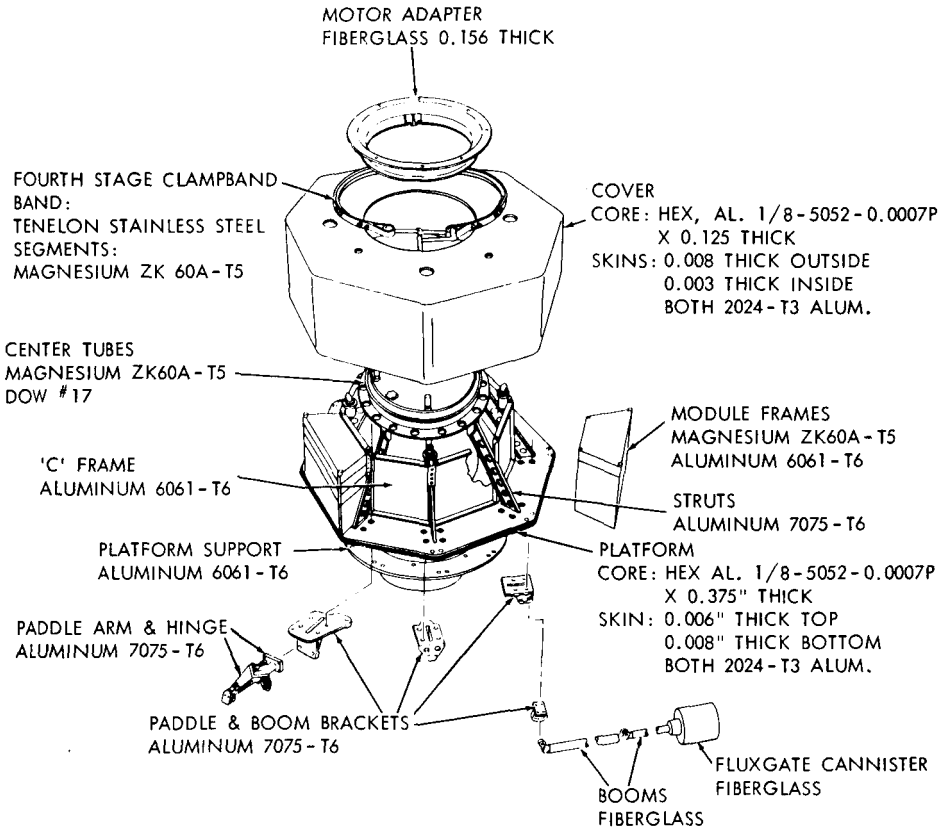

Figure 6. AIMP-E Spacecraft Showing Spacecraft Configuration and Structural Materials

DECONTAMINATION AND SAMPLING PROCEDURE

Electronic Circuit Modules

The electronic modules were the first component parts of the spacecraft that were decontaminated. Decontamination was achieved by first precleaning with an aerosol of ethyl alcohol to remove deposits of solder flux and/or water lacquer remaining on circuit boards after their fabrication. A control strip was then affixed to the module frame. This control strip was first sterilized in a steam autoclave before attachment. The modules were then inspected and electrical tests as required by the cognizant scientist were conducted. The modules were then delivered to the Mechanical Systems Branch at Goddard for bio-monitoring and integration into the spacecraft system.

A coupon was taken from the control strip that was affixed to the module and an assay performed to determine the type and level of contamination. The module and the remaining portion of the control strip were then decontaminated

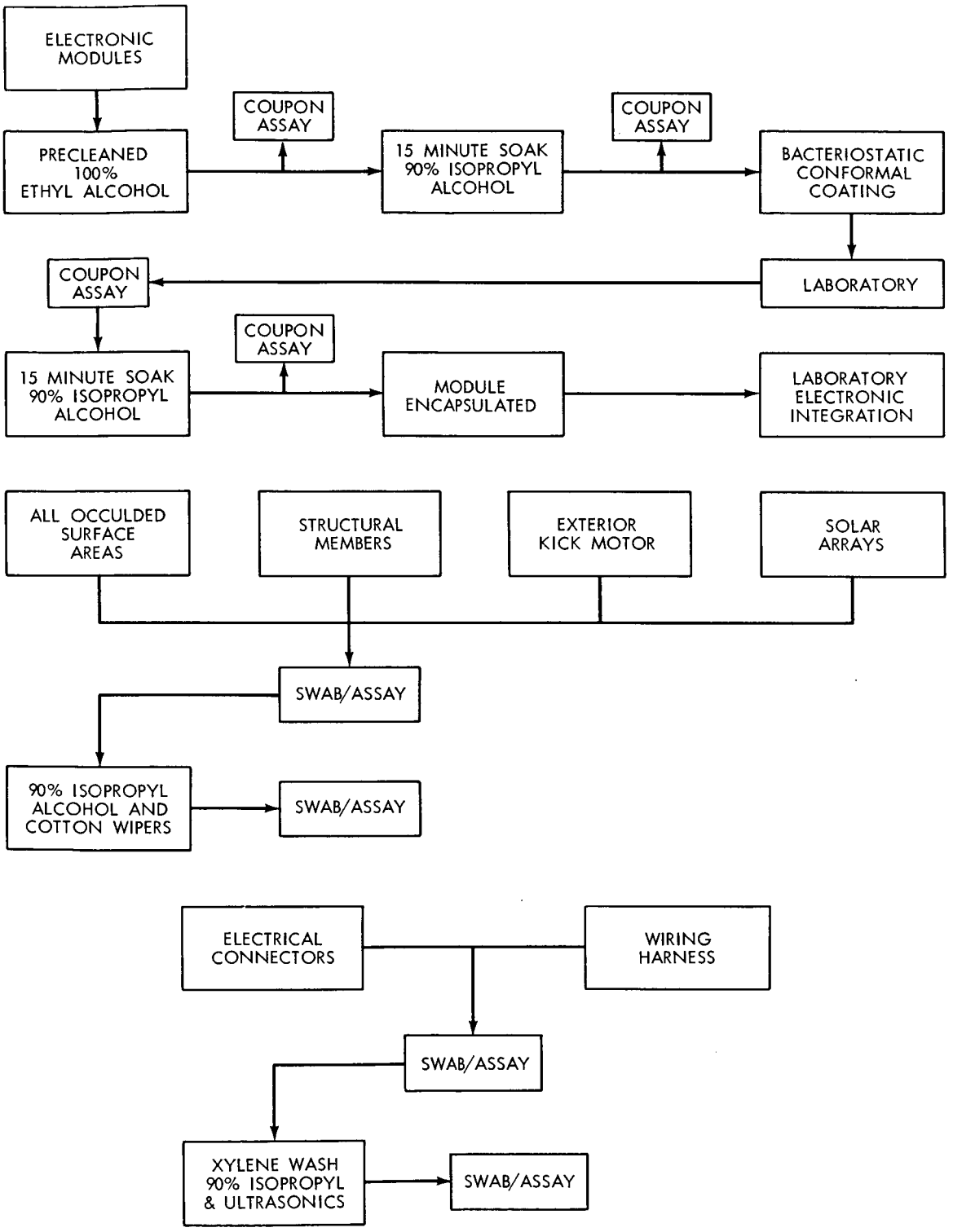

Figure 7a. Areas Decontaminated, Manner of Decontamination and Sterilization, and the Method of Recovering Viable Microorganisms

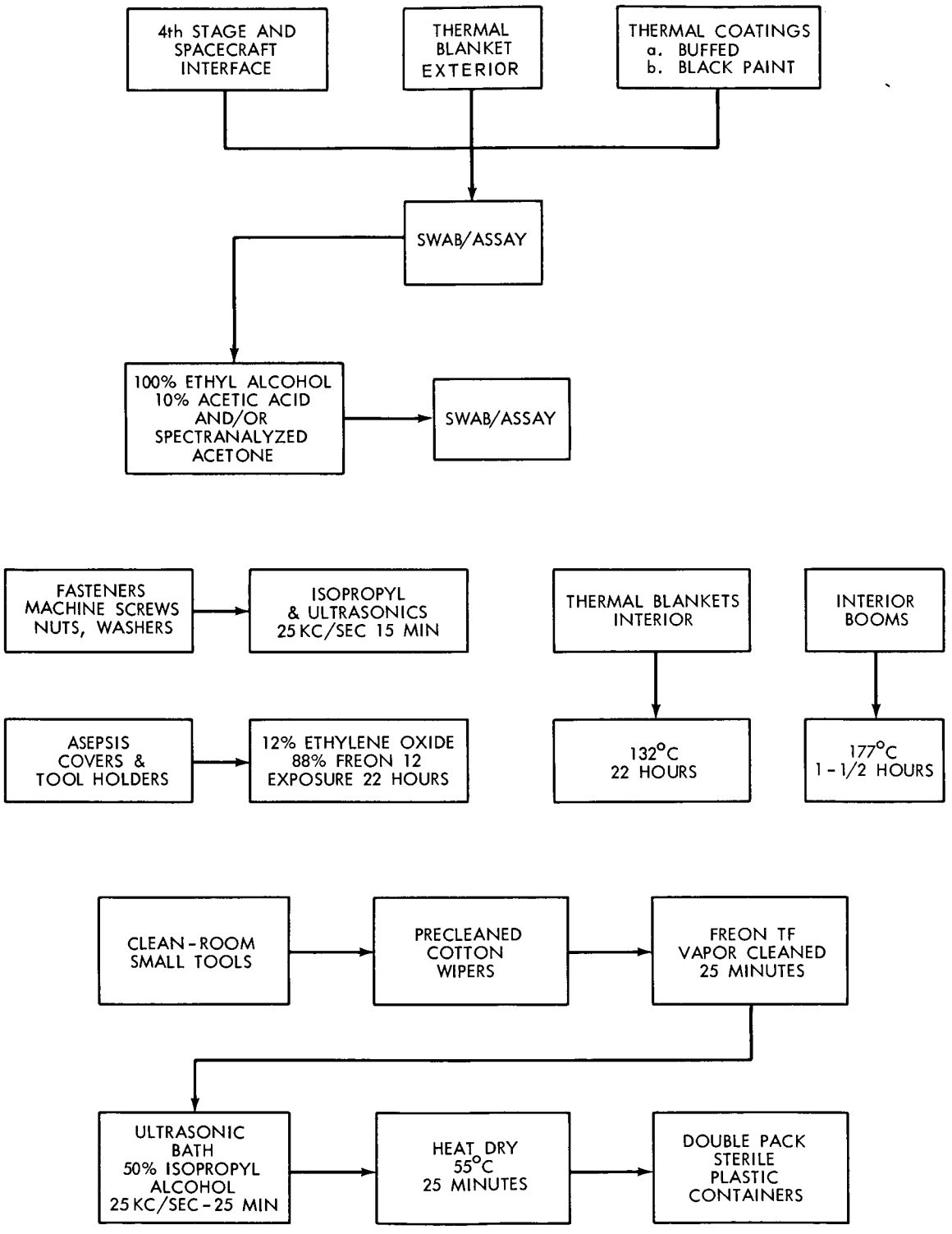

Figure 7b. Areas Decontaminated, Manner of Decontamination and Sterilization, and the Method of Recovering Viable Microorganisms

by immersion in a 90% isopropyl alcohol solution. The module was agitated by hand at least three times during a 15-minute immersion. The electronic circuit module and remaining portion of control strip was then placed into a vacuum oven and subjected to a temperature of 55°C for one hour duration. A coupon was then removed aseptically and an assay performed to determine the type and level of contamination remaining on the module after decontamination process. A bacteriostatic conformal coating was then applied to the electronic circuits and module. Board coating had bacteriostatic qualities on the following organisms: Bacillus subtilis var niger, Staphylococcus aureus, Pseudomonas alcaligines and Corynebacterium SSP.

The circuit modules were then packaged into a protective container and delivered to the laboratory for electronic checks and testing. Upon completion of the testing the circuit modules were sent to the Mechanical Systems Branch and the above process was repeated with the exception of the conformal coating. At this time the circuit modules were encapsulated after decontamination and assaying.

Occluded Surfaces; Structural Members and Motor Exterior

Prior to occlusion by a component, a structural member or attachments such as the retromotor, the area that was to be occluded was first assayed to determine contamination level. Sterile lint-free cotton wipers and isopropyl alcohol was used to effect decontamination.

The areas to be decontaminated were first precleaned by vacuuming the area to remove any gross particles. The cotton wipers were then immersed into a 90% solution of isopropyl alcohol, and wrung damp dry by hand. The pertinent areas were then thoroughly wiped. After evaporation of alcohol, a swab sample was taken. The number of samples taken was dependent upon the area of item sampled. The larger the area the greater number of samples were taken.

Electrical Connectors and Wiring Harness

The electrical connectors and wiring harness had the rosen flux deposits removed from the connections by applying xylene with a natural bristle brush for approximately fifteen seconds. Connectors were then rinsed off in an isopropyl bath. A swab sample was taken in several areas of the wiring harness for assays to determine the contamination level. The entire wiring harness including connections was then immersed in a 90% isopropyl bath and sonicated at 40 kc/sec for approximately ten minutes. Harness was then allowed to air dry under a fume hood and swab samples taken for determining levels of contamination after decontamination.

Interface, Thermal Blankets and Coatings

The fourth stage interface, thermal blanket exterior surface and the black painted and buffed thermal coatings were decontaminated in a like manner. Prior to any decontamination a swab sample was taken for assaying the contaminated level just prior to launch. After sample was taken any stubborn stain remaining on the coatings was removed with a 10% acetic acid solution and/or swabbed with a 100% ethyl alcohol. The entire surface area was then swabbed with sterile cotton swabs that had just been immersed in acetone. Only the purest grade of spectanalyzed acetone was used. A swab sample was then taken for assay of the decontaminated surfaces. After samples were taken that particular area was again swabbed with acetone.

Thermal Blankets and Interior of Booms

The thermal blankets were first assayed with swabs to determine the contaminated level. Samples were taken from several of the layers that made up the blanket. The blankets were then sterilized with dry heat, 140°C for 16 hours. After installation on spacecraft the exposed exterior surfaces were again monitored to determine the contamination level. The blankets were sealed; therefore it was accepted that sterility of the inner surfaces was not violated. The interior of the booms were considered sterile as they were subjected to 177°C dry heat for a period of 1 and 1/2 hours during lacquer bake-on. However, the exterior of booms and thermal blankets were exposed after sterilization and were subject to possible contamination, therefore they were again sampled, decontaminated and sampled to determine contamination levels just prior to launching spacecraft.

Miscellaneous Small Metallic Parts, Nuts, Bolts, Screws and Washers

The small miscellaneous metallic components were monitored for contamination by immersing 25% of their total number in a wash solution and plating out aliquots of solution as per the previously described technique. Decontamination of subject items was accomplished by immersion in 90% isopropyl alcohol for 15 minutes. The items were then placed into a 1% peptone wash solution and sonicated for 12 minutes at 25 kc/seconds. Samples of the solution were then plated out.

Asepsis Covers and Tool Holders

The asepsis covers for the retromotor, booms and entire spacecraft were wrapped individually in Kraft paper and inserted in a plastic container. They were then subjected to an environment of 12% ethylene oxide and 88% Freon 12

at ambient temperature of 72°F. The items were exposed at a slight positive pressure for a period of 22 hours.

Clean Room Small Tools

Tools used in the clean room during spacecraft assembly and/or field testing of the system were first precleaned by wiping off gross contamination with cotton wipers. Tools were then placed into a wire mesh basket and exposed to Freon TF* vapor cleaning for 25 minutes. They were then placed in an ultrasonic bath containing a 50% solution of isopropyl alcohol (C_3H_7OH) and sonicated for 25 minutes at 25 kc/sec. After removal from the solution they were placed in an oven which was preheated to 55°C. They remained in this environment for 25 minutes. All tools were then packaged and sealed in sterile plastic sheet material. These packs were again packaged so as to have tools double packed and sealed. The outer package was removed just prior to injecting tools into the Goddard Down Flow Unit for use in spacecraft assembly. To prove or disprove the adequacy of the above decontamination procedures tests were conducted on metal plates 4 x 4 square inches. These squares were left unattended and handled by several individuals so as to contaminate them as would be done in conducting an operation with tools manufactured from like materials. Dirty samples were first taken and assayed so as to determine the type and levels of contamination prior to decontamination. Swab samples were also taken after Freon Vapor Cleaning (FVC) and after the exposure to the 55°C heat drying. The test samples were placed in plastic containers and allowed to remain therein for 72 hours prior to final biosampling. The 72 hour period was selected to simulate the normal incubation period. Test samples were handled aseptically during all operations. Assays under aerobic conditions were performed to determine the reduction of micro-organisms that were achieved. The entire process was repeated with assays performed under anaerobic conditions. Table 1 depicts the results of these tests. It is to be noted that zero counts were obtained after each step in the decontamination process.

Assembly Environment (Figure 8)

It is felt that the greatest source of contamination to a spacecraft system will be from the technicians themselves and from the generation of debris that occurs during the spacecraft build-up, mechanical integration and/or final assembly, and checkout of flight configuration in the field. It was therefore determined that adequate clean-room facilities should be procured that would allow under aseptic conditions decontamination of spacecraft and its components, biosampling of areas for assays, mechanical integration, final assembly and tests.

*TRI-CHLORO-MONO-FLUORO-METHANE

Table 1

Condition	Contaminated				Decontaminated			
	Aerobic		Anaerobic		Aerobic		Anaerobic	
	Veg	Spores	Veg	Spores	Veg	Spores	Veg	Spores
1. Steel	4	0	4	0	0	0	0	0
2. Brass	139	0	27	0	0	0	0	0
3. Aluminum	2	0	1.0	0	0	0	0	0
4. Copper	0	0	0	0	0	0	0	0

Spacecraft Preparation Area

This was a controlled area where the debris generating operations were performed on a component or the spacecraft structure. When it was required to custom fit a component to the structure the operations of filing, drilling or scraping of metal were necessary a shield was built to protect items not worked upon from falling metallic particles. In addition a vacuum cleaner was employed to gather loose chips as generated. The inlet of vacuum cleaner was placed in the immediate area worked upon. The spacecraft or component was again vacuum cleaned before leaving this area.

Hi-Bay Clean Room Complex (Figure 9)

The Hi-bay clean-room complex consisted of a 100,000 class conventional clean-room of approximately 70 feet square, 24 feet high. Within this clean-room are class 100 portable Vertical Laminar Flow Units which are expandable in multiples of 4 x 8 units and Class 100 horizontal flow benches. The vertical flow units were used to house the spacecraft when not worked upon in the class 100,000 area. The spacecraft was precleaned each time before placing it under the down flow units. Cleaning consisted of wiping and vacuuming the surfaces. The down flow units were also used to perform instrument integration, decontamination and bio-sampling of components. The flow benches were used when assembling delicate mechanisms and the cleaning thereof at each stage of assembly. After the spacecraft was mechanically integrated it left the clean room area for electronic integration and/or systems environmental tests. The spacecraft was protected at that time by a strippable coating which was applied only to the exterior exposed surfaces.

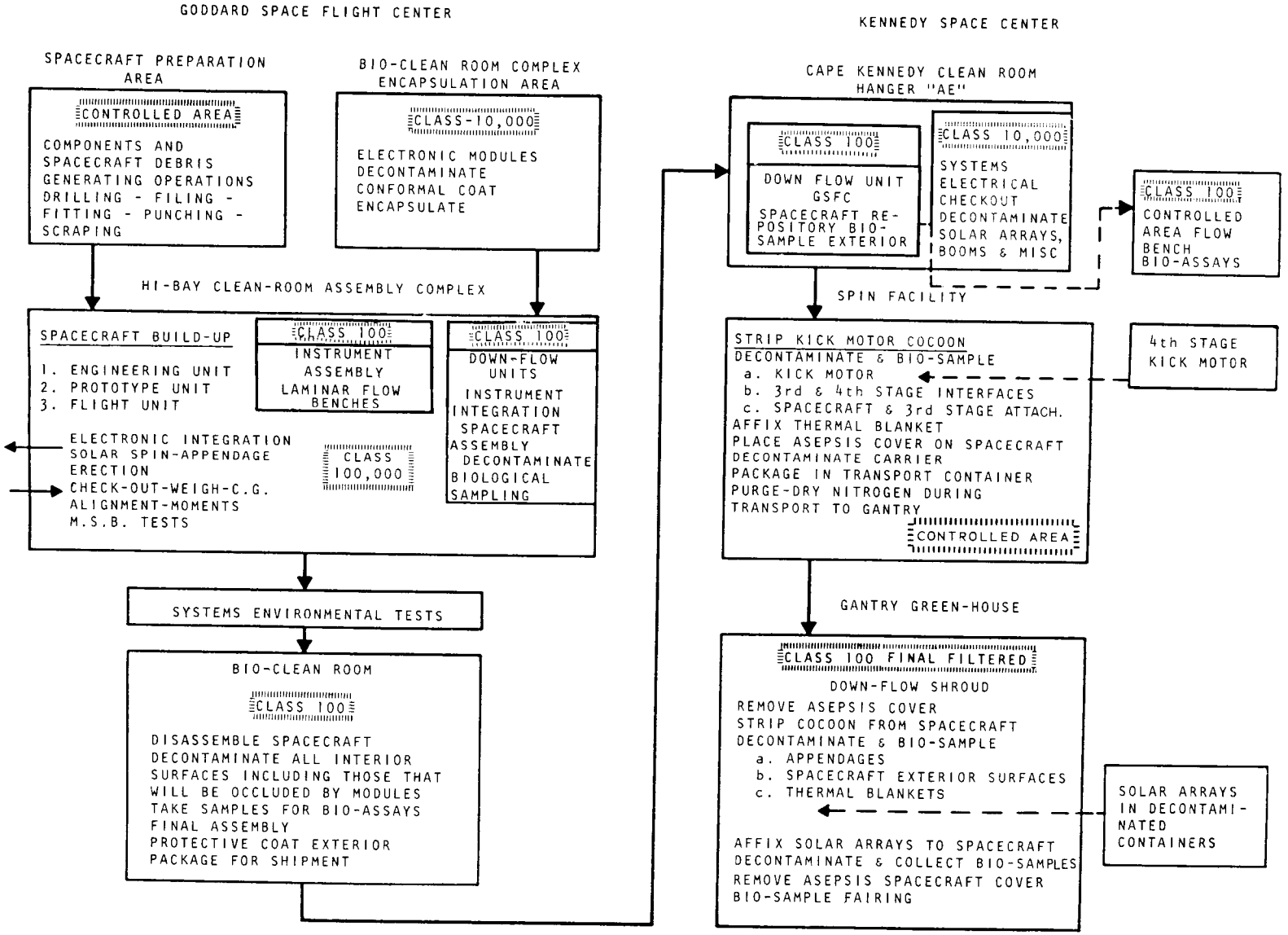

Figure 8. Assembly Environment

Figure 9. Hi-Bay Clean Room Complex

Bio-Clean Room (Figure 10)

Room D is a bio-clean room where satellites are decontaminated. An unusual feature of this room is a monitoring camera which photographs the satellite and personnel every 5 seconds. This feature was included to check on faulty operations that may occur while in the bio-clean room. Horizontal, laminar-flow air emanates from a 14-foot wall via modules with Cambridge high efficiency particulate air (HEPA) filter units. Filtration tests confirmed a rating for this room between 0 and 66 particles of 0.5 micron or less per cubic foot of air. Walls and ceiling are of prefabricated panels with a 4 inch insulation of plastic foam. Epoxy-coated steel forms the interior surfaces. A completely lighted ceiling gives a shadowless, 200-foot candle illumination at working levels. There are a minimum of 20 air changes per hour at a temperature range of 67°F to 77°F and a relative humidity level of 40 to 45 percent. A constant temperature of 72°F was maintained. A central built-in wall-type vacuum system is provided in all four rooms, along with observation windows that are double-paned and sealed. Also included are pass-through chambers containing interlock doors to assure maintenance of a positive air-pressure when parts are brought into the room.

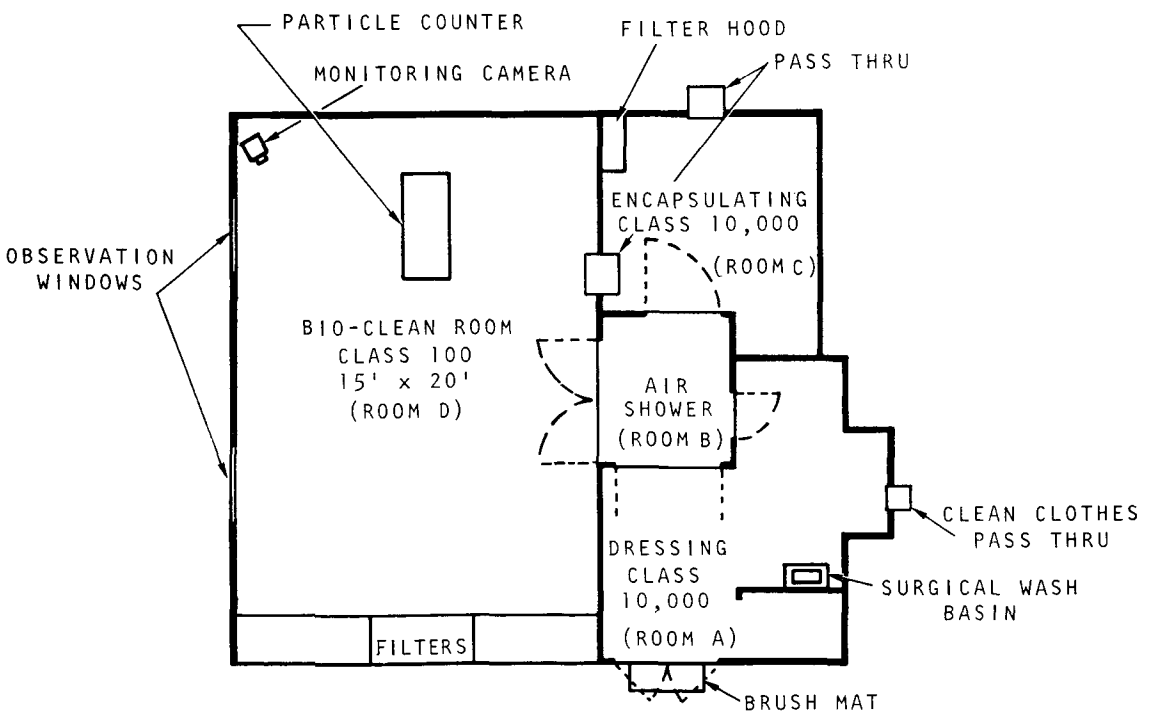

Figure 10. Bio-Clean Room

Upon returning from the systems environmental tests the spacecraft was returned to the Mechanical Systems Branch for disassembly, decontamination, bio-sampling and reassembly. These tasks were conducted in the confines of the bio-clean room complex.

The spacecraft was first precleaned with a vacuum and then wiped down with lint-free cotton wipers that had been immersed in isopropyl alcohol and wrung damp dry. The top cover was taken off and all the flight electronic modules and some instruments removed. The electronic circuit modules and instruments were then transferred to the Class 10,000 area of the clean room complex. Swab samples were taken and assays performed for determination of contamination type and levels. These items were then decontaminated with isopropyl alcohol and cotton wipers. The decontaminated circuit modules and instruments were then passed aseptically into the Class 100 area of the complex. The micro-biologist then took swab samples of each item decontaminated.

The spacecraft (Figure 11) was placed upon a dolly that had previously been decontaminated and then moved into the Class 100 area of the complex. The spacecraft on its dolly remained approximately one foot from the face of the air inlet filter tank during final assembly. Personnel remained downstream of the spacecraft at all times.

The area that modules and instruments occluded was then bio-sampled, decontaminated and bio-sampled. Sampling was accomplished by swab method and decontamination accomplished with isopropyl alcohol dampened cotton wipers. The modules were then integrated into the spacecraft structure. After the integration was completed all the exposed surface areas on the interior of the spacecraft were bio-sampled for contamination type and level, decontaminated with isopropyl alcohol and then bio-sampled and assays performed to determine the number of organisms remaining after decontamination. The inside of the cover was treated in a like manner and then installed on the spacecraft. Bio-sampling and decontamination were achieved in the same manner as the exposed interior areas. The exterior of the top cover was then cleaned and decontaminated, however, bio-samples were not taken at this time. The exposed surface areas of the spacecraft were then coated with a strippable coating. The spacecraft was then placed into a container that had previously been decontaminated with isopropyl alcohol and cotton wipers. Spacecraft container was then flushed with GN_2 and pressurized to 15 psia with dry nitrogen. The spacecraft was then shipped by aircraft to the Kennedy Space Center for field tests and launch.

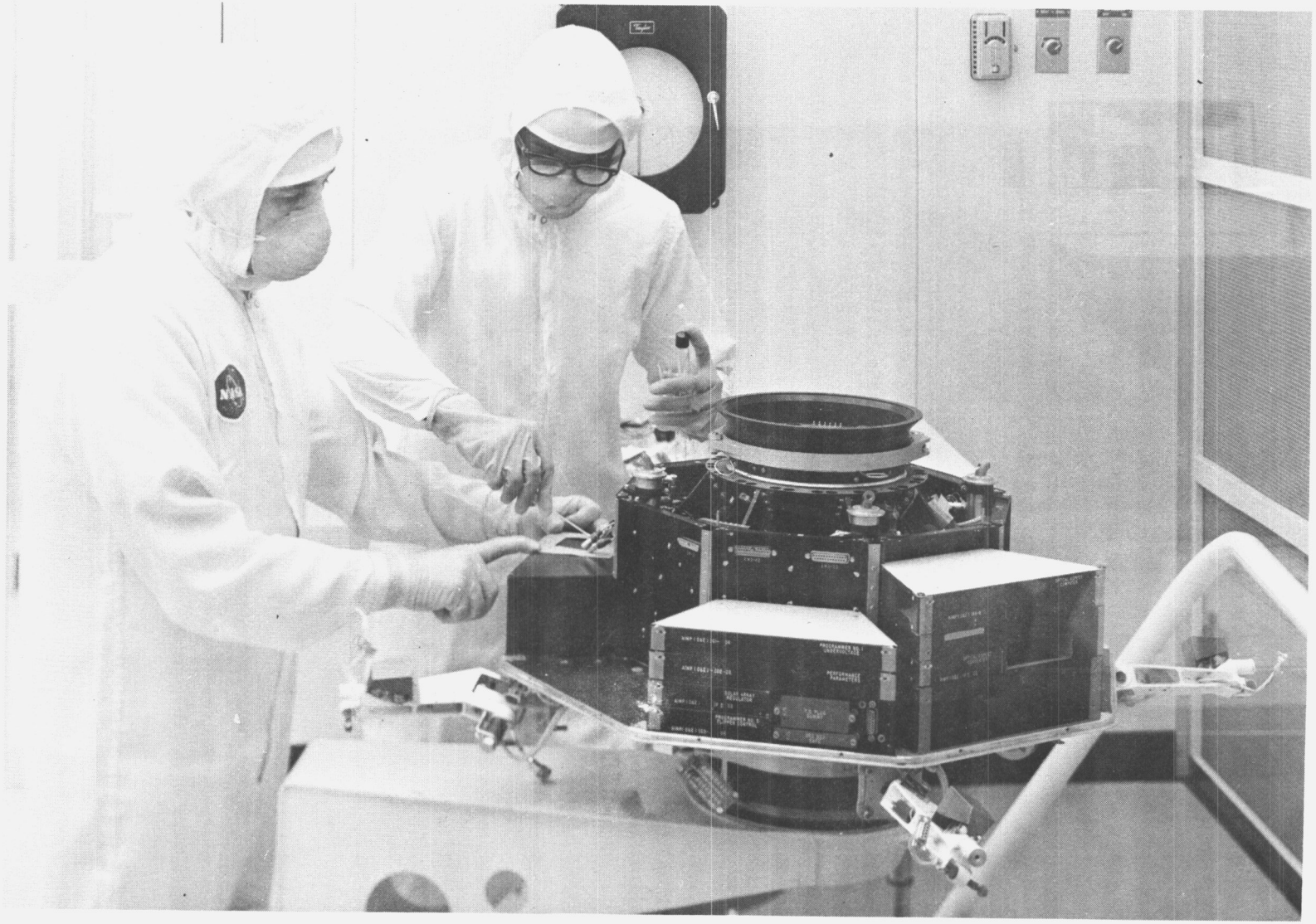

Figure 11. The Spacecraft During Final Assembly

ASEPSIS CONTROL DURING CONDUCTANCE OF FIELD TESTS (CAPE KENNEDY)

Sun-Spin Facility

After completion of preliminary operational checkout of spacecraft the body of the spacecraft was covered with a clean asepsis covering. The spacecraft was placed into its special container, transported to the sun-spin facility and then mounted upon the spin table. The solar paddles were then affixed to their supports. The asepsis covering remained on the spacecraft during all tests conducted in this area.

After completion of the sun-spin tests the solar paddles were removed. Swab samples were then taken on the solar paddles for assays to determine the bio-contamination type and levels in the contaminated state.

The spacecraft was packaged and then transported to the air-lock of the AE clean room.

Cape Kennedy Clean Room Facility (Figure 12)

The Cape Kennedy Class 10,000 clean room, located in the AE building, was used to house the Goddard Class 100 laminar down-flow unit.

The laminar downflow unit and the exterior of the spacecraft container were first decontaminated before moving them into the clean room.

The Goddard Down Flow Unit which housed the AIMP-D spacecraft during various tests and experiments was decontaminated and assembled in the airlock, Hi-bay clean room (Figure 9). The procedures employed include the following: decontaminating the spacecraft dolly which also involved removing the strip coating; preparation of ground support equipment (GSE) which entailed removing connecting cables, air inlet filters, all tape and paper units, and filling the voids in instrument racks with cover plates, and replacing air inlet filters with new filter material; and decontaminating electronic equipment connection cables by passing them between two sponges that were soaked in an alcohol solution. Mitocs, telephones, hand tools, and lead pigs that contained radioactive sources were treated in the same manner.

The spacecraft was removed from its container and placed upon the previously decontaminated dolly. The dolly and spacecraft were then placed under the down flow unit and remained there during the field checkout tests.

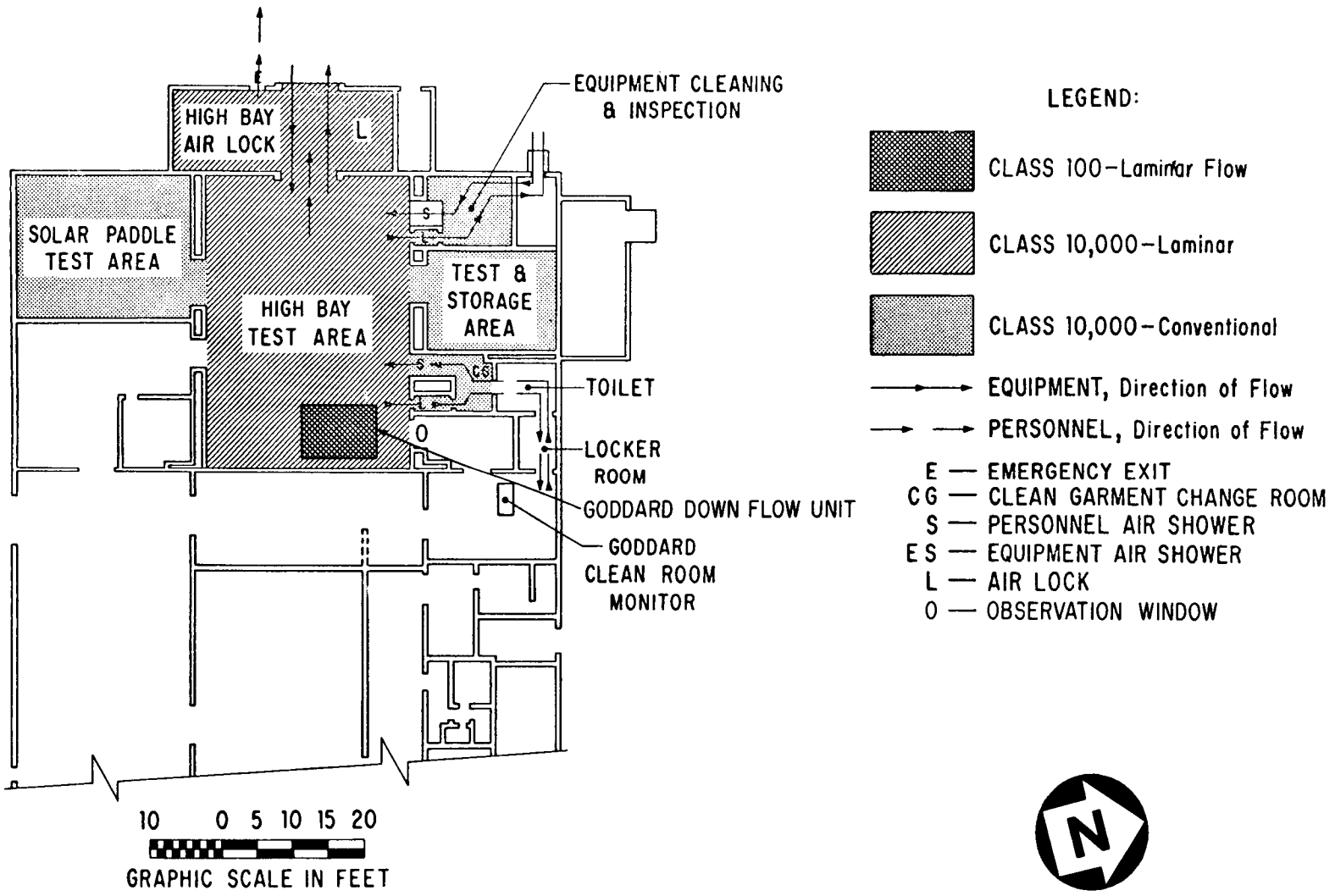

Figure 12. Spacecraft Clean Rooms, Building "AE"

Authorized persons who were conducting tests and experiments or working in any of the clean room areas were subjected to the following partial list of regulations:

- Personnel with respiratory malfunctions, skin ailments, colds, and severe sunburn were not permitted in clean room areas.
- Test fixtures, tools, jigs and assembly fixtures that were necessary to perform specific tasks were permitted.
- No abrasives, e.g., file, crocus cloth, etc., and no shredding or masking tapes.
- Exposed parts or components were never left on work benches.
- Approved clean room garments were worn in various clean room areas as previously mentioned.
- Smoking or eating was not permitted.
- Scratching of the head, eyebrows, and other exposed areas of the skin was forbidden.
- Only 5 persons were allowed in the Hi-Bay clean room or any of the other clean room areas at any one time.
- No more than two persons were allowed under the GSFC Down Flow Unit at the same time.
- Hand tools that were not in use were stored in a decontaminating solution.

The clean room regulations as stated above were necessary to assure asepsis handling of the AIMP spacecraft during experiment check-out phases.

Air Sampling

Trays of stainless steel strips (1 x 2 inches) were positioned on one side of the laminar crossflow, Class 10,000 clean room, upstream, downstream and midway of the clean room and one tray was placed in the Class 100 down flow unit bench high at working level. Six strips were recovered once a week from each location and assayed for aerobic and anaerobic vegetative and sporalitive viable life. Air samples were collected on two occasions; prior to introducing spacecraft into down flow unit and during field checkout testing. Renier slit

samplers with a one-hour clock were used. Air was drawn into the samples at a rate of 1 cubic foot per minute for 60 minutes. Glass plates (150 x 20 mm) containing 60 ml of Tripticase Soy Agar were used as collectors in the samplers. Air samples were placed approximately at bench top level (3 feet from the floor) in three different positions - upstream, downstream and in the Goddard down flow unit.

Table 2 lists the microbial contamination in the air of the clean rooms which housed the AIMP-E flight spacecraft during field checkout tests, as well as the number of personnel in the room during sampling period.

Table 3 lists the microbial fallout on the stainless steel strips over a 4-week period.

After completion of the electronic and instrument checkouts the spacecraft was removed from the Goddard laminar down flow unit and placed into its container while in the AE Class 10,000 clean room. The container had previously been decontaminated with alcohol and cotton wipers.

Spin Balance Facility

The spacecraft fourth stage and retromotor interfaces were swabbed sampled for assays to determine contaminated state. They were then decontaminated and swabbed sampled for assays to determine the bio-burden remaining after decontamination process. The thermal blanket that had previously been sterilized was then placed over the fourth stage retromotor. The third stage and spacecraft interfaces were treated in a like manner as the fourth stage interfaces. The solar paddles were then attached to the body of the spacecraft. The third and fourth stage vehicles and the spacecraft in its flight configuration were dynamically and statically balanced as a unit. After the balance tests the paddles were removed and transported back to the AE clean room for final decontamination and bio-assays.

An asepsis cover, previously sterilized with ethylene oxide compound, was placed over the entire spacecraft. The entire unit was then placed in a previously decontaminated transfer container. The container was sealed and was then pressurized with a slight positive pressure of gaseous dry nitrogen. The canned unit was then transported to the gantry and affixed to the second stage vehicle.

Gantry Operations

The transfer container was removed and the asepsis bag was allowed to remain intact over the spacecraft until the air cooling hat-shroud was placed in

Table 2

Microbial Contamination in the Air of Laminar Flow Clean Rooms Over a 7 Hour Period

Hour	Viable Particles Per Cubic Foot Per Hour					
	Crossflow Room Downstream		Crossflow Room Center		Crossflow Room Center	
	Date: 6/9/67		Date: 6/9/67		Date: 6/9/67	
	Count Part./ft^3	Personnel	Count Part./ft^3	Personnel	Count Part./ft^3	Personnel
1	0	0	0	0	0	0-1
2	0	0-2	0.033	0-2	0.050	0-2
3	0	0-1	0	0-1	0.033	0-1
4	0	0-2	0.016	0-2	0	0-1
5	0	0-1	0	0-1	0	0-3
6	0.016	0-1	0	0-1	0	0-2
7	0.083	0-1	0	0-1	0	0-2
Average Viable Particle/ft^3	0.0141		0.007		0.0119	

Microbial Contamination in the Air of Laminar Flow Clean Rooms Over a 4 Hour Period

Hour	Viable Particles Per Cubit Foot Per Hour					
	Downflow Downstream		Downflow Upstream		Crossflow	
	Date: 6/27/67		Date: 6/27/67		Date: 6/27/67	
	Count Part./ft^3	Personnel	Count Part./ft^3	Personnel	Count Part./ft^3	Personnel
1	0.166	0-2	—*	0-2	0.050	3-5
2	0.033	1	0.033	1	0.050	0-3
3	0	0-3	0.033	0-3	0.016	0-5
4	0.016	0-2	0	0-2	0.016	1-4
Average Viable Particle/ft^3	0.0537		0.0220		0.0330	

*Not Countable

Table 3
Microbial Fallout on Stainless Steel Strips

Viable Organisms Per Square Foot																
Week of Exposure	Downflow Room				Crossflow Room											
					Upstream				Center				Downstream			
	Vegetative Organisms		Spores		Vegetative Organisms		Spores		Vegetative Organisms		Spores		Vegetative Organisms		Spores	
	Aerobic	Anaerobic	Aerobic	Anaerobic	Aerobic	Anaerobic	Aerobic	Anaerobic	Aerobic	Anaerobic	Aerobic	Anaerobic	Aerobic	Anaerobic	Aerobic	Anaerobic
1	0	0	0	0	4	6	0	0	3	0	0	0	25	2	0	0
2	1	1	2	3	5	4	7	1	6	6	4	2	2	2	3	0
3	3	0	0	0	8	0	3	0	0	0	8	0	3	0	2	0
4	0	1	0	0	-	-	-	-	-	-	-	-	-	-	-	-

operation. The cooling hat shroud was first LOX compatible cleaned and decontaminated with isopropyl alcohol before assembly on the gantry. Verification that the filtered air supplied to the cooling hat shroud was Class 100 was authenticated by chemical engineers from PanAm using a Royco particle counter. Three tests were run. One test for one minute and two runs of 10 minutes each. The filtered air entered from the top of the cooling shroud. It was temperature controlled and passed through a diffuser designed to assimilate a vertical laminar flow of air over the spacecraft. The sides of the cooling hat shroud were rolled up to permit mating of the nose cone fairing which was fitted to the Delta vehicle. During this operation the asepsis cover remained on the spacecraft to protect it from fall-out of particulate matter. During this operation the flow of air was continued. The sides of the shroud were let down after the fitting.

The nose cone fairing was decontaminated and packaged prior to delivery to gantry and/or assembly on vehicle.

The solar paddles were decontaminated in the Goddard down flow unit, placed into containers that were previously decontaminated with isopropyl alcohol. Prior to launch the asepsis covers were removed and all the protective strippable coatings were removed. All exposed surfaces were then swab sampled for bio-assays in contaminated state.

The thermal coatings were decontaminated with triple-distilled acetone, spectranalyzed grade. However, all stubborn deposits were first removed with a 10% acetic acid solution and 100% ethyl alcohol. All other surfaces were decontaminated with a 90% solution of isopropyl alcohol. Prior to installation of the solar paddles, all mating surfaces were decontaminated with isopropyl alcohol prior to occlusion. Tie-down cords were then affixed.

All representative areas were biologically sampled so that assays could be performed to determine burden at time of launch.

The asepsis cover was replaced on the spacecraft. Nose cone fairing asepsis covering was then removed.

One-half of the nose cone fairing that was previously decontaminated was mated to the vehicle. The filtered conditioned air umbilical was connected to this section of nose cone fairing. The spacecraft asepsis cover was removed just prior to mating the other half of the nose cone fairing.

The spacecraft was constantly bathed with Class 100 filtered and temperature conditioned air until separation of umbilical at time of lift-off.

RESULTS

On the basis of the Goddard Bio-Records (see Table 4), it was determined that the surfaces of the AIMP-E spacecraft contained not in excess of 9×10^5 microorganisms prior to decontamination, and not in excess of 2.7×10^4 microorganisms after the decontamination process. This constituted a 97% reduction of organisms. The estimation of viable internal burden of components (see Table 5) was based upon past history and known manufacturing environments. It was determined that, of the total viable life remaining in the components, (see Table 7) 10% would be spore forms. Of this 10% approximately two-thirds would be aerobic and the remainder anaerobic.

As a result of the overall evaluation (see Table 6), it was determined that at time of launch the AIMP-E spacecraft contained not in excess of 2.5×10^5 organisms. Of these an estimated 2.2×10^5 organisms were contained inside the components and foam encapsulant, and 2.7×10^4 organisms on the surfaces, 7.4×10^3 of these were spores.

The AIMP-E spacecraft achieved a successful orbit with a life expectancy of three years and will have 1440 cycles temperature change between -45°C and +50°C in an ultra-high vacuum. Under this environment the spore population on the exposed surfaces of the spacecraft should be reduced to 1.89×10^{-9} at time of lunar impact, and all vegetative life assumed no longer to exist, only the components internal spore burden (2.2×10^4) would remain.

The Planetary Quarantine Officer, NASA Headquarters, recommended certification of the AIMP-E spacecraft based upon the evaluation of records maintained at the Goddard Space Flight Center, visual observations of control procedures, and assessment of the microbial environment of the spacecraft while in residence at the Eastern Test Range.

Table 4

Area Class	Total Area (in^2)	Compilation of Counts of Viable Organisms on Surfaces							
		Contaminated				Decontaminated			
		Aerobic		Anaerobic		Aerobic		Anaerobic	
		Veg.	Spores	Veg.	Spores	Veg.	Spores	Veg.	Spores
Occluded "A": Electronic Modules	8759	212899	62232	26858	25547	5937	3251	4099	55
Occluded "B": Surfaces that Module Frames Occlude	1138	4033	0	5914	0	72	0	3450	0
Occluded "C": Exterior of Module Frames	5813	56300	4489	9923	85	20	2030	192	0
Occluded "C": Other Interior Exposed Surfaces of the Spacecraft that the Cover Occludes	5195	20001	17467	898	40206	79	178	0	369
Interior Surfaces "D": Other Interior Surfaces of the Spacecraft —1 Body —2 Motor —3 Assembly Occluded	8490	63174	6517	8747	1750	1985	502	38	12
Exterior Surfaces "E": Exterior Surfaces of the Spacecraft —1 Body —2 Motor —3 Assembly Occluded	71437	228329	80818	19321	4344	3671	420	281	545
Final Totals of Contamination of the AIMP-E Spacecraft	Total 700.2 sq.ft.	584736	171523	71661	71932	11764	6381	8060	981
GRAND TOTALS		9×10^5				2.7×10^4			

Table 5

Compilation of Viable Organisms Contained Within Components

Components	Estimated Range	Number of Components	Accumulative Total x 10^3	
			Low	High
Resistors	0-1	11612	0	11.6
Capacitors	10-100	3153	31.5	326.9
Diodes	0-1	4005	31.5	330.9
Transistors	0-1	3164	31.5	334.1
Relays	100-1000	15	33.0	349.1
Crystals	0-1	1	33.0	349.1
Inductors	0 < 100	148	33.0	363.0
Toroids Transformers	0 < 100	117	33.0	375.6
Batteries	0	0	33.0	375.6
Metals	0	0	33.0	375.6
Tubes	0	4	33.0	375.6
Explosives	10	8	33.1	375.7
Foam	1/ml	14727 ml	47.8	390.4
Nylon-Dacron	0	876		
Teflon Insulation	0	16		
Magnetic Cores	0	0		
MOSFETS	0	747		
Pots	?	17		
Flat Paks	0	551		
Fuses	0	15		
Thermistors	0	35		
Estimated Total - Internal Burden			47.8	390.4
Average Internal Burden			219.0	

Table 6

Microbial Load at Launch AIMP-E Spacecraft	
Type Load	Contamination Level
Internal Burden	2.2×10^5
Surfaces	2.7×10^4
Total Load	2.5×10^5

Table 7

Estimated Spore Loading at Launch & Lunar Impact			
Area	Aerobic	Anaerobic	Totals
Surfaces	1.3×10^4	1.9×10^3	1.5×10^4
Internal Burden	1.5×10^4	7.3×10^3	2.2×10^4
Grand Totals	2.8×10^4	9.2×10^3	3.7×10^4
Remaining at Lunar Impact	1.89×10^{-9}		2.2×10^4